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Mr Brian McCaslin
FOLEY & LARDNER
Suite 500
3000 K Street, N.W.
WASHINGTON, D.C. 20007-5109

Paris, july 20th 1999

Dear Mr McCaslin,

Pleased find enclosed the signed declaration.

I look forward to hearing from you and remain.

Yours sincerely,

L. Chatenoud M.D., D.Sc.

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IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Attorney Docket No. 040388/0110

In re patent application of
Jean-Francois BACH *et al.*
Serial No. 08/986,568
Filed: December 5, 1997

Group Art Unit: 1644
Examiner: F. VanderVegt

For: METHOD FOR TREATING ESTABLISHED SPONTANEOUS AUTO-IMMUNE DISEASES IN MAMMALS

DECLARATION UNDER 37 C.F.R. § 1.132

Assistant Commissioner for Patents
Washington, D.C. 20231

Sir:

I, Lucienne Chatenoud, am a named inventor in the above-captioned application, and I declare as follows:

I. CREDENTIALS

I received a Medical Doctorate in 1979 from the Università degli Studi (Milano, Italy), and I then trained in Paris, as a nephrologist, at the Université Paris V. Concurrently, starting in 1980, I worked at the Hospital Necker in Paris, where I commenced my training in immunology under Professor Jean-Francois Bach. In 1986 I received a Doctorate in Science, in immunology, from the Université Paris VII. Currently, I am a research immunologist, under appointment by INSERM. I have more than 15 years of experience with the clinical and experimental use, in the context of autoimmunity and transplantation, of anti-T cell monoclonal antibodies. In this regard, I possess particular expertise relating to anti-CD3 antibodies.

II. THE SUBJECT APPLICATION DESCRIBES A PERMANENT STATE OF ANTIGEN SPECIFIC UNRESPONSIVENESS THAT IS NOT THE RESULT OF ANERGY INDUCTION

I have discussed with counsel the interview with Examiner VanderVegt, conducted on June 2, 1999, and I have reviewed the corresponding Interview Summary.

I understand that the Examiner expresses concern that there is no difference "between the permanent state of [u]nresponsiveness of the instant method and the 'two-signal' type hypothesis of anergy induction." See Interview Summary, June 2, 1999, Paper Number 10.

In addressing this concern (see below), I conclude that the methodology claimed in the subject application effects a permanent state of antigen specific unresponsiveness that is not related to the "two-signal" type hypothesis of anergy induction. Briefly, animals treated in accordance with the claimed method do not respond to exogenous interleukin-2 (IL-2), contrary to what one would expect if anergic cells were responsible for the observed unresponsiveness.

According to classical definition, anergic cells express IL-2 receptors and can be reverted to full responsiveness, both *in vitro* and *in vivo*, by adequate IL-2 supply. See Desilva *et al.*, *J. Immunol.* 147: 3261 (1991); Dallman *et al.*, *J. Exp. Med.* 173: 79 (1991). Furthermore, Desilva *et al.* have shown that anergic states of unresponsiveness induced by administration of the 145-2C11 anti-CD3 mAb, the same anti-body evaluated in the present invention, are reversed in response to exogenous IL-2. Thus, it would be expected, were anergic cells responsible for the observed antigen specific unresponsiveness of the present invention, that the resultant, induced protection would be reversed by an exogenous supply of IL-2.

As discussed in Chatenoud *et al.*, we tested for the presence of anergic cells in tolerant recipients, by attempting to reverse the anti-CD3 mAb-induced protection by an exogenous supply of IL-2. See Reference A1: Chatenoud *et al.*, *J. Immunol.* 1997: 2947. Murine P815 cells, which were transected with the mouse IL-2 encoding gene and which secreted the biologically active cytokine, were implanted *s.c.* into euglycemic NOD females, 8 to 10 weeks after anti-CD3 mAb treatment. In this manner, an exogenous supply of IL-2 was ensured for 7 to 8 consecutive days. None of the studied animals, however, showed relapse of IDDM, indicating that the remission

of diabetes, induced by treatment with anti-CD3 compounds, operated via a mechanism other than promotion of anergy.

Also, a durable state of unresponsiveness caused by an inhibition of lymphocyte functional capacity, itself a direct effect of the antibody or antibody fragment, then would be expected in all models, upon administration of an anti-CD3 compound. This supposition has proven false, however. It is well established, for example, that administration of anti-CD3 compounds does not effect the long-term survival rate of allograft transplantation recipients. See Hendrickson *et al.*, *Transplantation* 60: 828 (1995). In this model, therapies utilizing anti-CD3 compounds have induced transient immunosuppression only.

In light of the foregoing, I conclude that the permanent state of antigen specific unresponsiveness induced by the method claimed in the subject application is unrelated to the "two-signal" hypothesis of anergy induction.

I declare further that all statements made herein of my knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements and the like are made with knowledge that willful false statement and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of this application or any patent issuing thereon.

July 20th 1999
Date

Lucienne Chatenoud